

CLAIMS

What is claimed is:

5

1. A method for detecting the presence or absence of a bacterium, comprising the steps of:
 - a) contacting a sample with a detectably labeled synthetic serpin reactive site loop domain peptide substrate under conditions that result in modification of said substrate by an enzyme produced by a bacterium; and
 - b) detecting a modification or an absence of the modification of the substrate, the modification of the substrate indicating the presence of the bacterium in the sample and absence of the modification of the substrate indicating absence of the bacterium in the sample.

10

2. A method according to Claim 1, wherein the bacterium is a wound-specific bacterium selected from the group consisting of *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Streptococcus pyogenes*, *Pseudomonas aeruginosa*, *Enterococcus faecalis*, *Serratia marcescens*, *Proteus mirabilis*, *Enterobacter clocae*, *Acetinobacter anitratus*, *Klebsiella pneumonia*, and *Escherichia coli*.

15

3. A method according to Claims 1 or 2, wherein the enzyme is a protease.

20

4. A method according to any of Claims 1-3, wherein the substrate is labeled with a fluorescent probe and a quencher dye molecule.

25

5. A method according to any of Claims 1-4, wherein the substrate is labeled by a label selected from the group consisting of spin labels, antigen tags, epitope tags, haptens, enzyme labels, prosthetic groups, fluorescent materials, pH-sensitive materials, chemiluminescent materials, colorimetric components, bioluminescent materials, and radioactive materials.

30

6. A method according to any of Claims 1-5, wherein the substrate comprises at least one of the peptides selected from the group consisting of EAAGAMFLEAIPK, EGAMFLEAIPMSIPK,
5 KGTEAAGAMFLEAIPMSIPPEVK, GAMFLEAIPMSIPPE, and CGAMFLEAIPMSIPAAAHHHHH.
7. A method according to any of Claims 1-6, wherein the sample is selected from the group consisting of a wound surface on a subject and a body fluid.
10
8. A method according to any of Claims 1-7, wherein the substrate is on a solid support.
15
9. A method according to any of Claims 1-8, wherein the solid support is selected from the group consisting of a wound dressing, a container for holding body fluids, a disk, a scope, a filter, a lens, a foam, a cloth, a paper, a suture, a dipstick, a swab, a urine collection bag, a blood collection bag, a plasma collection bag, a test tube, a catheter, and a well of a microplate.
20
10. A method according to any of Claims 1-9, wherein the solid support comprises a material required to be free of microbial contaminants.
25
11. A method according to any of Claims 1-10, wherein the substrate comprises at least two dissimilar colorimetric components and the substrate is attached to a solid support, wherein modification of the substrate comprises cleaving at least a portion of the substrate that includes one of the colorimetric components, the cleaving resulting in a visible color change.
30
12. A method according to any of Claims 1-11, wherein the colorimetric components are covalently attached to the peptide.
13. A biosensor for detecting the presence or absence of a bacterium in a sample, the biosensor comprising:

- a) a solid support and
- b) a detectably labeled synthetic serpin reactive site loop (RSL) domain peptide substrate, said substrate attached to said solid support.

5

14. A biosensor according to Claim 13, wherein the substrate is specific to a protein produced by a wound-specific bacterium selected from the group consisting of *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Streptococcus pyogenes*, *Pseudomonas aeruginosa*, *Enterococcus faecalis*, *Serratia marcescens*, *Proteus mirabilis*, *Enterobacter clocae*, *Acetinobacter anitratus*, *Klebsiella pneumonia*, and *Escherichia coli*.

10

15. A biosensor according to Claims 14, wherein the protein is a protease enzyme

15

16. A biosensor according to any of Claims 13-15, wherein the substrate is labeled with a fluorescent probe and a quencher dye molecule.

20

17. A biosensor according to any of Claims 13-16, wherein the substrate is labeled by a label selected from the group consisting of spin labels, antigen tags, epitope tags, haptens, enzyme labels, prosthetic groups, fluorescent materials, pH-sensitive materials, chemiluminescent materials, colorimetric components, bioluminescent materials, and radioactive materials.

25

18. A biosensor according to any of Claims 13-17, wherein the substrate comprises at least one of the peptides selected from the group consisting of EAAGAMFLEAIPK, EGAMFLEAIPMSIPK, KGTEAAAGAMFLEAIPMSIPPEVK, GAMFLEAIPMSIPPE, and CGAMFLEAIPMSIPAAHHHHH.

30

19. A biosensor according to any of Claims 13-18, wherein the solid support is selected from the group consisting of a wound dressing, a container for holding body fluids, a disk, a scope, a filter, a lens, a foam, a cloth, a paper, a

suture, a dipstick, a swab, a urine collection bag, a blood collection bag, a plasma collection bag, a test tube, a catheter, and a well of a microplate.

20. A biosensor according to any of Claims 13-19, wherein the solid support
5 comprises a material required to be free of microbial contaminants.

21. A biosensor according to any of Claims 13-20, wherein the substrate
comprises at least two dissimilar colorimetric components covalently
attached to the peptide.

10 22. An isolated peptide comprising a detectable label and an amino acid
sequence selected from the group consisting of EAAGAMFLEAIPK,
EGAMFLEAIPMSIPK, KGTEAAGAMFLEAIPMSIPPEVK,
GAMFLEAIPMSIPPE, and CGAMFLEAIPMSIPAAHHHHH.

15